ajava

Asian Journal of Animal and Veterinary Advances

Academic
Journals Inc.

www.academicjournals.com

Asian Journal of Animal and Veterinary Advances 8 (2): 284-292, 2013
ISSN 1683-9919 / DOI: 10.3923/ajava.2013.284.292

Performance of Commercial Poultry Offal Meal as Fishmeal Replacement in the Diet of Juvenile Malaysian Mahseer, *Tor tambroides*

Saufinas Ismail, Mohd Salleh Kamarudin and Ehsan Ramezani-Fard
Department of Aquaculture, Faculty of Agriculture, Universiti Putra Malaysia, 43400 Serdang, Selangor, Malaysia

Corresponding Author: Mohd Salleh Kamarudin, Department of Aquaculture, Faculty of Agriculture, Universiti Putra Malaysia, 43400 Serdang, Selangor, Malaysia

ABSTRACT

Mahseer is one of the most expensive freshwater fish in Malaysia and has a good potential for freshwater aquaculture industry. This study was conducted to evaluate the suitability of Poultry Offal Meal (POM) as Fish Meal (FM) replacement in the diet of Malaysian mahseer (*Tor tambroides*). The POM:FM ratios tested were 0:100, 25:75, 50:50, 75:25 and 100:0. One hundred and fifty mahseer fingerlings (3.0±0.05 g) were equally and randomly stocked into 15 65 L aquaria. The diets were randomly assigned to the aquaria in triplicates groups. The feeding trial was conducted for 96 days. No significant differences ($p>0.05$) were observed among the survival rate of fish while it was above 90% in all the treatments. Weight gain and specific growth rate significantly increased when 50-100% of dietary fishmeal was substituted with POM. Whole body composition of Malaysian mahseer was not significantly influenced by the experimental diets. This study showed that fishmeal in the diet of mahseer can be replaced by POM up to 100% ratio without any adverse effect on survival, growth performance and body composition.

Key words: *Tor tambroides*, poultry offal meal, fish meal replacement, body composition, growth performance

INTRODUCTION

Mahseers (*Tor* spp.) are an important group of big scale riverine cyprinids occurring in mountainous rivers and lakes of most trans-Himalayan countries (Ramezani-Fard *et al*., 2011a). Its name may be derived from Hindi (mahasir: maha-great and sir-head), Sanskrit (maha-salka: large-scaled) or Indo-Persian (mahisher: mahi-fish and sher-tiger) (Thomas, 1897). In India, mahseer is known as the king of Indian freshwater systems (Nandeesha *et al*., 1993). In Malaysia, mahseer is one of the most expensive freshwater fish and has a good potential for freshwater aquaculture industry (Ramezani-Fard *et al*., 2011b; Misieng *et al*., 2011). There are three species of mahseer in Malaysia: *Tor tambroides*, *T. douronensis* and *T. tambra* (Ingram *et al*., 2005; Nguyen *et al*., 2006). *Tor tambroides* (Malaysian mahseer) has also a high demand in recreational and ornamental fish industry due to its character and attractive coloration (Ng, 2004). The distribution and abundance of this species have declined over the recent years due to significant degradation of their natural habitats caused by deforestation, overfishing and agricultural development (Ismail *et al*., 2011).

Ng *et al.* (2008) found that Malaysian mahseer fingerling needs about 45-50% dietary protein while Misieng *et al.* (2011) reported a lower optimum dietary protein requirement of 40%. Fishmeal and shrimp meal has long been used as a major protein source in aqua-feeds because of their high nutritional value (Siraj *et al.*, 1988; Hlophe and Moyo, 2011). Fish meal is of good quality protein and has an attractive odour which increases the palatability of fish diet (Adeniji, 2008). However, the availability of this feedstuff in future can no longer be assured because the ocean stocks for some species have been reduced (Hlophe *et al.*, 2011). Demand for fishmeal is still so high and is not commensurate with its production. The higher biological value of fishmeal compared to the other protein sources makes it more expensive than the others (Omole *et al.*, 2008). The high fishmeal price leads to the increase of feed cost (Hu *et al.*, 2008; Millamena, 2002). Therefore, finding an alternative protein source which can fully or partially replace fishmeal is now becoming increasingly important especially for fish such as mahseer that require high dietary protein (Ng *et al.*, 2008). Cruz-Suarez *et al.* (2007) reported that 50 to 65% replacement of fishmeal will reduce feed cost by 10 to 14% which in turn reduces the aquaculture production cost.

Several authors have studied the performance of Poultry Offal Meal (POM) in various fish species and observed that POM as a cheaper and good protein source can be comparable to fishmeal (Shapawi *et al.*, 2007; Emre *et al.*, 2003). POM in some countries is defined as a meal which is produced from viscera, heads and blood while in US it is defined as a meal containing all by-products of poultry including feathers (Dale *et al.*, 1993). POM has slightly higher in crude protein and fat compared to fishmeal (Omole *et al.*, 2008).

To date, there is no published information on the feasibility of using POM in mahseer diet as a fishmeal replacement. This study was conducted to determine the effects of replacement of dietary fishmeal with POM on the growth, survival and body composition of Malaysian mahseer and to determine the optimal replacement ratio of POM in the diet of this fish.

MATERIALS AND METHODS

Diet preparation: Five isonitrogenous and isocaloric diets were formulated in order to substitute different ratio of fishmeal (0 -control-, 25, 50, 75 and 100%) with POM (Table 1). The formulation

Table 1: The composition of test diets (% as fed basis)

Ingredient	Diet 1 (0:100)	Diet 2 (25:75)	Diet 3 (50:50)	Diet 4 (75:25)	Diet 5 (100:0)
Fish meal	40.00	30.00	20.00	10.00	0.00
Poultry offal meal	0.00	10.00	20.00	30.00	40.00
Soybean meal	28.19	28.65	29.12	29.61	30.11
Corn meal	0.00	0.00	0.09	0.45	0.81
Rice bran	1.55	2.28	2.91	3.23	3.55
Tapioca starch	20.00	20.00	20.00	20.00	20.00
Vitamin premix	1.00	1.00	1.00	1.00	1.00
Mineral premix	1.00	1.00	1.00	1.00	1.00
Vegetable oil	8.26	7.07	5.88	4.71	3.53
Proximate composition					
Moisture (%)	10.35	10.35	10.40	10.05	10.30
Crude protein (% DM)	32.04	34.02	33.76	33.58	37.25
Crude fat (% DM)	8.43	9.36	6.86	7.65	7.19
Crude fiber (% DM)	1.48	2.28	1.52	1.61	1.87
NFE (% DM)	32.59	34.80	37.35	38.44	35.62
Ash (% DM)	15.10	9.18	10.12	8.68	8.17
Gross energy (kJ g^{-1})	16.90	17.80	18.00	18.10	18.00

NFE: Nitrogen-free extract

was performed using the winfeed 2.8 for Windows (Winfeed Limited, Cambridge, UK). All diets contained 35% crude protein and 17.5 kJ g^{-1} gross energy (Misieng *et al*., 2011). The diets were pelletized using a single screw extruder (Brabender KE19, die size 3 mm Ø). The barrel temperatures were set at 60-100-120°C and the temperature at the die head was 160°C. The feeding, shaft and four-bladed cutter speeds were set at 50, 150 and 310 rpm, respectively. The extruded pellets were stored in sealed plastic bags at 4°C until use.

Feeding trial: The feeding trial was conducted at the Aquaculture Experimental Station, Universiti Putra Malaysia. *Tor tambroides* fry were procured from a local fish supplier and acclimatised in a 1 ton PE tank for 7 days. During the acclimation period, fish were fed diet 1 (Table 1). One hundred and fifty fingerlings (3.0±0.5 g) were equally and randomly assigned to 15 glass aquaria with capacity of 65 L. Continuous filtration and aeration kept dissolved oxygen level in each aquarium above 5.5 mg L^{-1}. Water temperature, pH, dissolved oxygen and ammonia nitrogen were monitored twice per week. Water temperature was found between 23 and 3°C while pH ranged between 6.5 and 8.8. The toxic ammonia (NH_3^+) was below 0.01 mg L^{-1}. The experiment was conducted for 14 weeks from May to August 2011 and fish were fed twice per day (09:00 and 18:00 h) at 3% b.wt. Fish were sampled and weighed every two weeks and the quantity of feed was adjusted accordingly. Aquaria water was partially changed (70%) during each sampling. Dead fish were collected and sent to Universiti Putra Malaysia Veterinary Hospital in order to diagnose causes of death. At the end of the experiment, Weight Gain (WG), Specific Growth Rate (SGR), Feed Conversion Ratio (FCR) and Protein Efficiency Ratio (PER) were calculated using following formulae. The fish were then sacrificed and stored at -45°C for further whole body proximate analysis:

$$\text{WG (\%)} = \frac{\text{Final weight (g)} - \text{Initial weight (g)}}{\text{Initial wieght (g)}} \times 100$$

$$\text{SGR} = \frac{\text{In final mean weight} - \text{In initial mean weight}}{\text{Experimental days}} \times 100$$

$$\text{FCR} = \frac{\text{Feed fed (g)}}{\text{Gain in weight of fish (g)}} \times 100$$

$$\text{PER} = \frac{\text{Increment in body weight (g)}}{\text{Protein intake (g)}} \times 100$$

Chemical analysis: The proximate analyses of the diets and fish were carried out according to AOAC methods (AOAC, 1997). The proximate analyses were done in triplicates. Whole fish samples were prepared through freeze-drying and grinding. Moisture content was estimated by drying samples in an oven at 105°C for 24 h to constant weight. The crude protein (nitrogen×6.25) was determined by the Kjeldahl method while crude lipid was estimated by ether extraction using a Soxhlet system. The crude fibre was determined using acid/base digestion followed by burning the samples to ash in a muffle furnace at 600°C for 12 h. The gross energy of diets and fish was determined by direct combustion in an adiabatic bomb calorimeter.

Statistical analysis: All data were subjected to one-way Analysis of Variance (ANOVA) using SPSS 17 for Windows (SPSS INC., Chicago, IL, USA) and the difference was considered significant at $p<0.05$. Duncan's Multiple Range Test was used to test mean differences. All percentage data were arcsine transformed prior to statistical analyses.

RESULTS

The survival rate of *T. tambroides* was generally high (90-100%) and no significant differences ($p>0.05$) among treatments were observed (Table 2). Few fish on diet 1 and diet 3 died because they were accidently trapped and killed by the filter. Few fish on diet 2 also died in the last two weeks due to non-infectious problems. However, a non-pathogenic or opportunistic group of bacteria (*Klebsiella* sp.) in fish were isolated from them.

The highest percentage of weight gain was observed in fish fed diets 3, 4 and 5 and there were no significant differences ($p>0.05$) between fish fed these diets (Table 2). However, weight gain in fish fed diets 1 and 2 were significantly lower ($p<0.05$) than those fed the other diets. Unlike the weight growth pattern, a rapid length growth was observed among all treatments in the first two weeks. However, the rate of length growth was slowly decreased after this period. Final total length in fish fed diets 3, 4 and 5 were significantly higher ($p<0.05$) than those fed diet 1 and 2. The highest total length gain was observed in fish fed diet 4 (31.60±2.63%) while it was not significantly different ($p>0.05$) with those fed diets 3 and 5.

The specific growth rate was generally low and showed a similar pattern as the weight gain. Fish on diet 4 showed the highest SGR while it was not significantly different ($p>0.05$) with those fed diets 3 and 5 and was significantly higher than fish fed diets 1 and 2 ($p<0.05$). Feed Conversion Ratios (FCR) of the experimental diets are shown in Table 2. The lowest and the best FCR (3.27±0.20) was achieved when fish fed diet 4 while it was not significantly different with fish fed diets 3 and 5. The FCR in fish fed diets 1 and 2 were significantly higher ($p<0.05$). The best Protein Efficient Ratio (PER) was also observed in fish fed diets 3, 4 and 5.

Table 3 shows the whole body composition of Malaysian mahseer juveniles fed different diets. Protein (39.37-45.65% DM) and crude fat (42.61-47.94% DM) were the main components of whole body of the fish. No significant differences ($p>0.05$) were observed among the whole body composition of fish fed different diets.

Table 2: Survival rate, growth performance and feed efficiency of Malaysian mahseer fed diets containing different percentage of POM as fishmeal replacement

Parameters	Diet 1 (0:100)	Diet 2 (25:75)	Diet 3 (50:50)	Diet 4 (75:25)	Diet 5 (100:0)
Survival rate (%)	93.30 ± 3.30^{a}	90.00 ± 10.0^{a}	96.70 ± 3.30^{a}	100.00 ± 0.00^{a}	100.00 ± 0.00^{a}
Final body weight (g)	9.90 ± 0.40^{a}	10.00 ± 0.50^{a}	11.40 ± 0.40^{b}	13.10 ± 0.50^{c}	12.40 ± 0.20^{bc}
Weight gain (g)	2.70 ± 0.20^{ab}	2.60 ± 0.30^{a}	4.00 ± 0.50^{bc}	5.20 ± 0.40^{c}	4.70 ± 0.30^{c}
Weight gain (%)	39.00 ± 0.80^{a}	33.30 ± 3.70^{a}	53.90 ± 7.70^{b}	66.80 ± 5.20^{b}	60.90 ± 4.50^{b}
Final length (cm)	9.36 ± 0.03^{ab}	9.29 ± 0.16^{a}	9.49 ± 0.13^{abc}	9.87 ± 0.20^{c}	9.82 ± 0.14^{bc}
Length gain (%)	24.80 ± 0.40^{ab}	23.90 ± 2.20^{a}	26.50 ± 1.70^{abc}	31.60 ± 2.60^{c}	31.00 ± 1.90^{bc}
SGR (% day^{-1})	0.34 ± 0.01^{ab}	0.29 ± 0.03^{a}	0.44 ± 0.05^{bc}	0.52 ± 0.06^{c}	0.48 ± 0.05^{c}
FCR	5.43 ± 0.31^{b}	5.79 ± 0.28^{b}	3.52 ± 0.19^{a}	3.27 ± 0.20^{a}	3.51 ± 0.23^{a}
PER	0.08 ± 0.00^{ab}	0.08 ± 0.01^{a}	0.11 ± 0.01^{bc}	0.15 ± 0.01^{c}	0.13 ± 0.01^{c}

Values within the same row having the same superscript are not significantly different at $p>0.05$, SGR: Specific growth rate, FCR: Feed conversion ratio, PER: Protein efficiency ratio

Table 3: Whole body proximate composition of juvenile *Tor tambroides* before and after feeding the experimental diets

Parameters	Initial	Diet 1 (0:100)	Diet 2 (25:75)	Diet 3 (50:50)	Diet 4 (75:25)	Diet 5 (100:0)
Moisture (% WW)	74.45	63.63±0.88[a]	66.06±1.79[a]	62.43±1.02[a]	66.35±1.93[a]	65.65± 1.80[a]
Protein (% DM)	48.57	45.65±1.23[a]	44.53±3.38[a]	39.37±1.92[a]	41.97±0.72[a]	41.16±1.56[a]
Ash (% DM)	10.02	9.63±0.54[a]	9.77±0.66[a]	8.80±0.66[a]	8.14±0.52[a]	8.82±0.31[a]
Fat (% DM)	32.40	42.61±1.83[a]	43.16±1.42[a]	47.94±1.80[a]	44.10±2.49[a]	45.51±0.21[a]
Fiber (% DM)	0.70	0.46±0.28[a]	0.63±0.14[a]	0.60±0.06[a]	0.39±0.07[a]	0.62±0.16[a]
NFE (% DM)	3.83	1.73±2.55[a]	1.91±5.21[a]	3.30±1.01[a]	5.40±1.93[a]	3.90±1.13[a]

Values within the same row having the same superscript are not significantly different at $p>0.05$, NFE: Nitrogen free extract

DISCUSSION

Malaysian mahseer is a slow-growing species which takes a longer time to reach to marketable size compared to other carp species (Ingram *et al*., 2005; Ng *et al*., 2008). The specific growth rate of *T. tambroides* in this study (0.29-0.52% day^{-1}) was similar to those reported by Ramezani-Fard *et al*. (2012), Kamarudin *et al*. (2011) and Misieng *et al*. (2011). However, this rate was lower than the other *Tor* species. Indian mahseer *T. putitora* grows at 1.5-1.8% day^{-1} (Rahman *et al*., 2005).

A longer culture period definitely increases the production cost through the feed, labour and maintenance costs. In addition, Malaysian mahseer requires high dietary protein Ng *et al*. (2008) and Misieng *et al*. (2011) which in turn correlates with a higher feed price due to the use of more fishmeal as the main protein source in the feed. Fishmeal supply is very limited, so its price inflates as the demand goes up every year (Shapawi *et al*., 2007). To reduce the fishmeal dependency and feed price, fishmeal needs to be partially or fully replaced by other alternative protein sources. Alternative protein sources such as POM or poultry by product meal, soybean meal, animal protein mixture and bone and meat meal have been studied by several researchers (Yang *et al*., 2004; Hernandez *et al*., 2007; Wang *et al*., 2008; Adewolu *et al*., 2010). However, most of these alternatives can be considered as a partial substitute for fishmeal. POM is a protein source mainly used in pet foods because of its palatability, high quality protein and essential fatty acids, vitamins and minerals (Cruz-Suarez *et al*., 2007). A high quality POM contains about 70% crude protein and relatively low ash content (Nengas *et al*., 1999; Davis and Arnold, 2000) and it is suggested that this feed stuff can be partially or fully substituted for fishmeal (Ogale, 2002). Shapawi *et al*. (2007) expressed that POM has a high potential to be integrated in the diet of carnivorous fish species such as groupers. In agreement with our findings, Takagi *et al*. (2000) also suggested that POM can be used in fish diet without any significant depression in fish performance. It should be considered that 50% replacement of dietary fishmeal with POM will lead to a cost reduction of about 10-14% per ton of feed (Cruz-Suarez *et al*., 2007).

The FCR values of 3.27 to 5.79 have been recorded in this study. Misieng *et al*. (2011) observed better FCR values of 2.19 to 3.02 for *T. tambroides* while Ramezani-Fard *et al*. (2012) and Kamarudin *et al*. (2011) reported FCR values of 1.4-2.0. The range of FCR in well-prepared fish diets is generally between 1.2 and 1.5 (De Silva and Perera, 1985). However, the FCR trend in the present study was in agreement with the earlier research in which FCR in the diets of some fish such as gibel carp (Yang *et al*., 2006) and humpback grouper (Shapawi *et al*., 2007) decreases with increasing inclusion of POM. The PER found in this study (0.08-0.14) was slightly higher than that was reported for Malaysian mahseer by Misieng *et al*. (2011) [0.06-0.11]. High PER suggested that fish were able to digest the diets containing POM and to absorb their nutrients efficiently.

The study showed that fishmeal can be fully replaced by POM in the Malaysian mahseer diet without any adverse effect on its growth performance and feed utilization. Yang *et al.* (2006) reported that high quality poultry by Product meal (PBM) can fully replace fishmeal in the diet of gibel carp. However they recommended an optimal fishmeal replacement of only 66.5%. In contrast, Emre *et al.* (2003) and Zabihi *et al.* (2011) reported a reduction in the growth of mirror carp, *Cyprinus carpio* fingerlings with the increase of dietary PBM level. This is due to the limited amino acids and the low digestibility of feather connective tissue and skin content in PBM. However, POM used in this study did not contain any feather materials.

Earlier works indicated that only 50% of fishmeal can be replaced by poultry by product meal in the diets of rainbow trout (Steffens, 1994), *Clarias gariepinus* (Adewolu *et al.*, 2010) and prawns (Yang *et al.*, 2004) without affecting their growth. Usman *et al.* (2007) also reported that poultry offal silage meal can only replace up to 37% of fishmeal in the diet of tiger grouper, *Epinephelus fuscoguttatus* without any adverse effects on fish productivity.

This study showed that the body composition of *T. tambroides* was not affected by the replacement of fishmeal with POM. Similar findings regarding the effects of diets on the body composition of Malaysian mahseer have been reported. Misieng *et al.* (2011) noted that an increase in the dietary protein level of *T. tambroides* does not affect its body protein, fat and NFE but increases its body ash content. Ramezani-Fard *et al.* (2012) also reported that the body composition of *T. tambroides* does not change when fish are fed with diets containing low or high omega-3 and low or high saturated fatty acids. However, Kamarudin *et al.* (2011) found that dietary lipid sources affect the body fat content of *T. tambroides*. The increase of whole body fat content has been observed in rainbow trout (Steffens, 1994) and mirror carp (Emre *et al.*, 2003) when they are fed with POM diets. However, the negative trend occurs in mirror carp (Emre *et al.*, 2003; Zabihi *et al.*, 2011). In the other hand, no differences in whole body moisture and fat are found in gibel carp (Yang *et al.*, 2006) and humpback grouper (Shapawi *et al.*, 2007) when POM is included in their diets. However, the whole body protein and energy content in gibel carp fed POM diets are slightly higher than those fed fishmeal diet (Yang *et al.*, 2006). Omole *et al.* (2008) reported that fat content of POM is higher than fat content of some local fishmeal.

CONCLUSION

In the present study, it is concluded that poultry offal meal could fully replace fishmeal in the diet of Malaysian mahseer (*Tor tambroides*) fingerlings without affecting the survival rate, growth performance and whole body proximate composition.

ACKNOWLEDGMENTS

The authors would like to thank Dinding Soya and Multifeeds Sdn Bhd (Malaysia) for the supply of poultry offal meal. This project was funded by the Malaysian Government through E-Science grant no. 05-01-04-SF0209.

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